Grape leaf rust mite, *Calepitrimerus vitis* (Acari: Eriophyidae), a new pest of grapes in British Columbia

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ABSTRACT

The grape leaf rust mite, *Calepitrimerus vitis* (Nalepa), was first discovered in the interior of British Columbia in 2009 on grape leaves from a commercial vineyard north of Osoyoos. Bronzing of grape leaves confirmed to be caused by *C. vitis* in summer 2009 was followed by severely stunted shoots and distorted leaves in several vineyards in spring 2010. Numbers and lengths of shoots and fruit clusters were reduced significantly on vines infested with *C. vitis*. Earlier studies have shown that outbreaks of *C. vitis* result from pesticide sprays targeted to other pests that damage predator mite populations. Sprays of sulphur-based fungicides early in the season are the recommended method of control.

INTRODUCTION

Grape leaf rust mite, *Calepitrimerus vitis* (Nalepa) (Eriophyidae), is a host-specific pest of grapevines, *Vitis vinifera* L., (Anonymous 1968; Bernard *et al*. 2005; Walton *et al*. 2007) found in most grape-growing regions of the world, including Washington State since 2002 and Oregon since 2004 (Prischmann and James 2005; Walton *et al.* 2007). *Calepitrimerus vitis* has not previously been reported on grapevines in British Columbia (B.C.) and was not found during an extensive survey of vineyard pests conducted in the Okanagan and Similkameen valleys in 1972 (Madsen and Morgan 1975).

Calepitrimerus vitis has been considered an economic pest of grapes only during the past four decades, possibly the result of reduced use of sulphur-based fungicides that provide effective control (Anonymous 1968; Barnes 1970; James 2007) and from increased applications of pesticides that are harmful to predators that normally keep its numbers in check (Winkler *et al.* 1972; Bernard *et al.* 2005; Schreiner *et al*. 2014). Bronzing of grape leaves in late summer can appear significant but is not thought to affect the current year's growth or quality of the fruit at fall harvest (Anonymous 2005; Reinert 2006; James 2007). However, leaf bronzing is a good indicator of the potential for large overwintering rust mite populations to emerge the following spring and continue feeding, resulting in damage to the developing buds, shoots and leaves (Bernard *et al.* 2005; Prischmann and James 2005; James 2007). Significant economic injury can occur to grapes if these mites are not properly managed. Feeding of overwintered *C. vitis* in spring on developing buds and shoots results in what has been termed short shoot syndrome or reduced spring growth (Bernard *et al*. 2005; Walton *et al*. 2007; Schreiner *et al*. 2014), which is typified by severely stunted growth, shortened internodes, scarring of shoots, curled and distorted basal leaves, and reduced fruit set. Severe infestations can result in abortion of affected bunches and complete crop loss (Walton *et al.* 2007). The relationship of impaired and damaged spring growth of grapevines to *C. vitis* feeding is not always clear. Several other factors may also be responsible for restricted spring growth, such as heavy thrips (Thysanoptera) feeding, winter freeze and herbicide damage (Schreiner *et al.* 2014).

This paper documents the first confirmed discovery of *C. vitis* in the Okanagan Valley of B.C. and the results of subsequent surveys to assess *C. vitis* abundance and its impact

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on vine growth. Grape growing and wine production are important industries in the Okanagan Valley, with 8,060 acres currently planted in wine grapes, accounting for 84% of B.C.'s vineyard acreage (British Columbia Wine Institute 2015). In order to provide advice to growers on the best management practices for this emerging pest, we also present information on recommended methods of control developed elsewhere that mostly rely on early season applications of sulphur.

MATERIALS AND METHODS

Identification of Calepitrimerus vitis. Inspection on June 24, 2009, of grapevines at a commercial vineyard north of Osoyoos (49° 05' 23" N, 119° 30' 39" W) that had heavily bronzed leaves revealed the presence of eriophyid mites. Samples of these mites were preserved in 70% ethanol and sent for species verification to Dr. F. Beaulieu, Canadian National Collection of Insects, Arachnids and Nematodes, Agriculture and Agri-Food Canada, Ottawa, and to Dr. J. Amrine, West Virginia University.

2009 Summer Survey. A survey for *C. vitis* was conducted during June 24 to September 8, 2009, in twelve vineyard cultivar blocks in eight vineyards located throughout the southern half of the Okanagan Valley from Summerland and Naramata in the north to Osoyoos in the south (Figure 1), including three blocks (Osoyoos: Shiraz, Merlot and Cabernet Sauvignon) adjacent to the vineyard block originally found to be infested on June 24. Two of the original Osoyoos blocks and a heavily infested site in Naramata were sampled 2–3 times over the course of the field season. Leaf samples consisted of 10 randomly selected leaves from each cultivar block. As per the protocol of Walton *et al.* (2007), mites from a sample were transferred to a glass plate by means of a mite brushing machine (J. G. H. Edwards, Okanagan Falls, B.C.); the glass plate was previously covered with a thin film of soap to immobilize the mites. The glass plate was then placed on a grid and examined using a dissecting microscope for counting all mite species and developmental stages.

2010 Spring Bud Dissections. In early spring 2010, buds from four vineyards found to be infested with *C. vitis* in 2009 were dissected, and all mite species under individual outer bud scales were counted. Vineyard managers of three of the four sampled vineyards applied sulphur (Kumulus; 80% sulphur; 2.86–11.2 kg/ha; BASF) during the woolly bud stage of development as per the recommended method of control (Anonymous 1968; Bernard *et al*. 2005). Ten randomly selected canes from each sampled cultivar block were pruned above the third bud and temporarily placed in cold storage for no more than one week until the assessments. Mite counts were conducted on the most basal bud from each pruned cane. This procedure was conducted once in March before the sulphur sprays and again in April approximately two weeks after the sulphur sprays.

Damage Assessment. The effect of early season feeding by *C. vitis* on developing shoots and fruit clusters was determined in a block of Chardonnay grapes at Okanagan Falls that had one section of eight vine rows heavily infested with *C. vitis* and showing symptoms of spring feeding damage (i.e., stunted and scarred shoots, small distorted leaves, brown and shrunken fruit clusters, etc.). The two adjacent damaged and undamaged sections of the block had been managed in exactly the same manner, except that the vineyard manager had sprayed the heavily infested rows the previous year with an undisclosed insecticide to control leafhoppers. No information about the spray application was provided. The use of a recapture sprayer for the insecticide application resulted in a clear differentiation between rows with and without *C. vitis* feeding damage.

Shoot lengths and counts of *C. vitis*, phytophagous thrips, and predatory mites on stems and the basal leaf of 20 randomly selected shoots per row were assessed on May 18, 2010, from four of the damaged and four of the undamaged rows. Predatory mites and phytophagous thrips were not identified to species. Data were collected from the third to sixth row away from the dividing line between the adjoining damaged and undamaged sections. Pre-harvest data were collected from half (one arm) of each of 20 randomly selected vines per row from those same four rows, with numbers of shoots and clusters counted and cluster lengths measured on September 15, 2010.

Statistical analysis. Shoot lengths, mite and thrips counts, and pre-harvest data from damaged and undamaged vine rows were analyzed using one-way ANOVA. All mite and thrips count data were transformed ($\sqrt{X + 0.5}$) before analysis (Zar 2010). Statistical tests were performed using JMP Version 10 (SAS Institute Inc. 2013), with all statistical error rates at $\alpha = 0.05$.

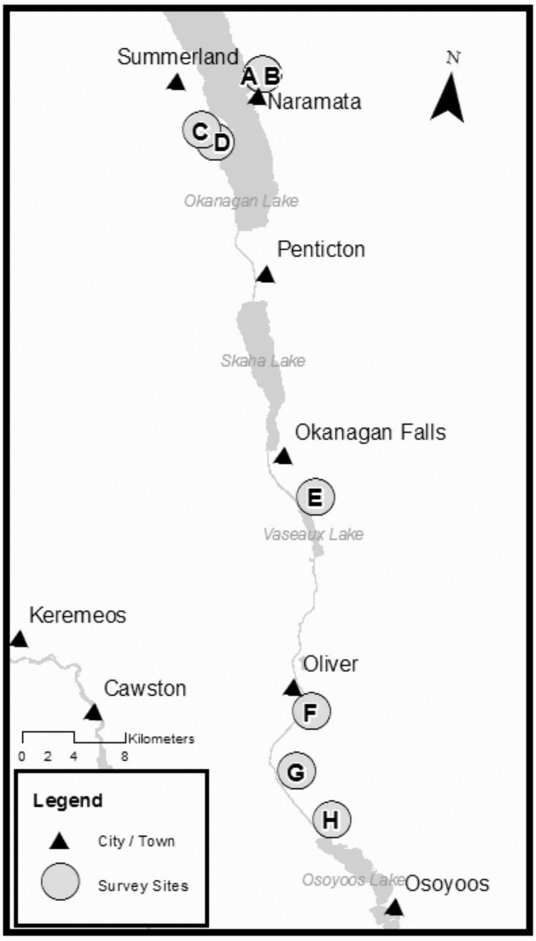

Figure 1. Locations of vineyards surveyed for *Calepitrimerus vitis* in the south Okanagan Valley, B.C. Counts of *C. vitis*, predatory mites, and tetranychid mites (numbers/leaf) for corresponding cultivars at each survey site are located in Table 1.

Table 1

Mean numbers of *Calepitrimerus vitis* [*Cv*], predatory mites [PM] and tetranychid mites [TM] per leaf from 12 cultivar blocks in eight vineyards in the Okanagan Valley, BC, sampled[a] between June 24 to September 8, 2009.

Map Label[b]	Location	Variety	Sample Date (2009)	*Cv*/leaf	PM/leaf	PM eggs/leaf	TM/leaf	TM eggs/leaf
A	Naramata	Merlot	Jul-07	269.2	0	0	7.2	15.2
A	"	"	Aug-05	998.0	1.8	1.4	11.0	12.4
B	Naramata	Pinot Gris	Jul-14	0	0	0	12.8	30.2
C	Summerland	Gewürztraminer	Jul-15	0	0	0	0	0
D	Summerland	Gewürztraminer	Sep-08	54.8	0	0	0	0
E	OK Falls	Pinot Noir	Jun-30	0	0	0	0.1	0
F	Oliver	Chardonnay	Jul-15	0	0	0	0	0
F	Oliver	Merlot	Jul-15	0	0	0	0.2	3.2
G	Oliver	Semillon	Jul-15	0	0	0	0.4	0.4
G	Oliver	Shiraz	Jul-15	0	0	0	0	0
H	Osoyoos	Cabernet Sauvignon	Jun-24	2.3	0	0	0.1	0.1
H	Osoyoos	Merlot	Jun-24	0.5	0	0	0	0
H	"	"	Sep-02	19.0	0	0	0.2	0.4
H	Osoyoos	Shiraz	Jun-24	139.6	0	0	0	2.0
H	"	"	Aug-05	191.0	0.4	0	11.0	21.2
H	"	"	Sep-02	55.2	0	0	4.0	6.6

[a]Mites removed from ten randomly selected leaves per block using a mite brushing machine.
[b]Letters correspond to vineyard locations on Figure 1.

RESULTS AND DISCUSSION

Identification of Grape Leaf Rust Mite. The eriophyid mites found to be the cause of bronzing or russeting of grape leaves in a commercial vineyard north of Osoyoos in the south Okanagan Valley in the summer of 2009 were confirmed by two independent experts to be *C. vitis* (see methods). This is the first record for B.C. Adults of this eriophyid mite species, a relative of the grape erineum mite, *Colomerus vitis* (Pagenstecher) (Eriophyidae), are approximately 0.15 mm long, light amber in colour, broader at the front end, and somewhat wormlike in appearance (Lowery 2015; Figures 2, 3). Feeding by *C. vitis* during the summer causes bronzing or stippling that can appear similar to spider mite (Tetranychidae) injury. Damage to leaves by *C. vitis* feeding is mostly restricted to the upper leaf surface rather than the lower surface and, unlike tetranychid mites, *C. vitis* does not produce webbing.

Figure 2. Adult grape leaf rust mite, *Calepitrimerus vitis* (Nalepa).

2009 Summer Survey. Of the eight vineyards sampled, *C. vitis* was found in several geographically separated locations (Table 1). Subsequent sampling has found it to be widespread in the valley with numbers as high as 998 mites per leaf. Numbers exceeding 3,000 per leaf have been recorded in Europe and Oregon on severely infested grapevines (Schreiner *et al*. 2014). The presence of *C. vitis* in multiple locations suggests that it has been present in south central B.C. for several years, but widespread movement by wind and with human activities (Duffner *et al.* 2001) could have resulted in rapid dispersal. Monitoring of *C. vitis* on three occasions in one of the surveyed blocks showed that numbers increased by early August and then declined in September (Table 1). Migration of *C. vitis* from leaves to their overwintering sites under outer bud scales and bark after the end of August agrees with reports by Walton *et al.* (2007) and Schreiner *et al.* (2014).

Grape varieties vary in their susceptibility to *C. vitis* feeding (Anonymous 2005; Bernard *et al.* 2005; Schreiner *et al.* 2014). Combined with cool spring temperatures, cultivars that develop slowly are exposed to the mites for a longer period and so damage may be more severe. Cabernet Sauvignon, a later developing cultivar, is reportedly more susceptible to *C. vitis* than earlier cultivars such as Chardonnay (Anonymous 2005), but our survey did not find large numbers of *C. vitis* on a Cabernet Sauvignon block adjacent to an infested Shiraz block (Table 1). Of the three cultivars sampled in the original vineyard, Shiraz leaves had the largest numbers of *C. vitis*. A Merlot block in Naramata also had high numbers of *C. vitis*. A larger study would be required to determine differences in susceptibility to *C. vitis* among cultivars under southern B.C. conditions.

There was no clear association between *C. vitis* numbers and numbers of tetranychid mites. The highest tetranychid counts (12.8/leaf and 30.2 eggs/leaf) were recorded in a vineyard (site B) where *C. vitis* was not found (Table 1). The second highest number of *C. vitis* (191) occurred on August 5 in a block of Shiraz (vineyard H) that initially had few tetranychids (2.0 eggs/leaf), but high tetranychid numbers were recorded from that same site in August and September. Although low numbers of *C. vitis* were most often associated with low numbers of tetranychids, the complexity of mite population dynamics combined with differing spray regimes confounds the relationship.

Figure 3. Overwintered adult female (deuterogyne) grape leaf rust mites, *Calepitrimerus vitis* (Nalepa), feeding under grapevine bud scales in spring.

2010 Spring Bud Dissections. *C. vitis* were found in large numbers in spring under the outer bud scales (Figure 3) of vines whose leaves had become bronzed the previous summer. Monitoring of eriophyid mites is difficult due to their microscopic size and cryptic nature (Schreiner *et al.* 2014). As an alternative to bud dissections, mite counts on double-sided sticky tape applied around the bases of developing shoots has been used to monitor *C. vitis* emergence in spring (Bernard *et al*. 2005), but the method was not found by Walton *et al.* (2007) to provide useful monitoring information. Scouting for signs of leaf bronzing or stippling in summer followed by an assessment of mite numbers provides a good indication of the need for control the following spring (Schreiner *et al*. 2014). As an alternative to a mite-brushing machine, Schreiner *et al.* (2014) developed a 'rinse in bag' system that extracted *C. vitis* from leaves into a small amount of ethanol or isopropanol for counting.

Sprays of sulphur-based materials during the woolly bud stage of grape development have been shown previously to effectively control *C. vitis* and prevent damage to developing shoots (Anonymous 1968; Bernard *et al*. 2005). Our dissections of buds in spring also indicated that sulphur (Kumulus™) applied by growers at the woolly bud stage was effective against *C. vitis*, as none were detected in the sprayed commercial vineyard blocks two weeks post-application (Table 2). For the cultivar block not sprayed with sulphur, *C. vitis* numbers increased ca. 29% over the same two-week time period. While not as effective, a single application of sulphur in mid-season was reported to reduce *C. vitis* populations by approximately 80% (Schreiner *et al.* 2014). Outbreaks of *C. vitis* in Washington State have been attributed to decreased use of sulphur for powdery mildew control (Prischmann and James 2005; Reinert 2006; James 2007). While reliance on sulphur sprays in the past for the control of fungal pathogens may also have provided control of *C. vitis*, high application rates can be detrimental to predacious mite populations (McMurtry *et al.* 1970; James 2007).

Predacious phytoseiid mites are known to provide effective control of eriophyid mites in the absence of insecticide sprays that are detrimental to their survival (James and Whitney 1993; Bernard *et al.* 2005). In the absence of sulphur sprays, conditions are right for *C. vitis* outbreaks following sprays that are harmful to mite predators. It was apparent, for example, that the application of an undisclosed insecticide to part of a cultivar block resulted in elevated numbers of *C. vitis* and significant damage to developing shoots (Figure 4) the following spring. Differences in *C. vitis* numbers between the heavily infested Shiraz block and the adjoining Merlot and Chardonnay blocks (Table 1) possibly reflects differing pesticide applications the preceding summer. Population levels of *C. vitis* would also vary depending on the frequency and timing of sulphur applications in spring.

Damage Assessment. *C. vitis* may have an uneven distribution even within a vineyard cultivar block. Assessment in spring 2010 of shoot growth in a section of a Chardonnay block that had been sprayed with an insecticide the previous summer showed severe stunting of shoots and distorted leaves (Figure 4). Differences in shoot lengths for the rows heavily infested with *C. vitis* were significantly shorter than those from the adjacent unsprayed rows ($F_{1,76}$=63.3543, P<0.0001) (Table 3). Examination of these damaged, or reduced, shoots showed significantly higher populations of *C. vitis* on the stems ($F_{1,76}$=76.5591, P<0.0001) and basal leaf ($F_{1,76}$=46.1787, P<0.0001) compared to undamaged shoots from the same vineyard block (Table 3). Numbers of phytophagous thrips were low, less than one per basal leaf or shoot, and were not found to differ significantly between the damaged and undamaged shoots ($F_{1,76}$=2.1008, P=0.1513) (Table 3); therefore, thrips are unlikely to have contributed to the damage. Injury was still measureable at harvest in September, with the *C. vitis*-infested vines having fewer shoots ($F_{1,78}$=5.5237, P=0.0213), fewer grape clusters ($F_{1,78}$=102.7369, P<0.0001), and shorter cluster lengths ($F_{1,78}$=18.3596, P<0.0001) (Table 3).

Table 2

Numbers (mean $\pm$ SE) of *Calepitrimerus vitis* (*Cv*) and predatory mites (PM) per grapevine bud scale recorded from four commercial vineyards. All vineyards were previously found to be infested with *C. vitis* in 2009 and were sprayed or not sprayed with Kumulus (80% sulphur) at the woolly bud stage in spring 2010 as part of individual growers' pest management programs. Mites were counted from under the outer bud scales of the third bud; canes were pruned above the second bud prior to the Kumulus spray and two weeks post-spray and temporarily stored at 2^{o} C.

Sulphur spray	Vineyard site (Map Label[a])	Cultivar	Date of pre-spray counts (2010)	Pre-spray counts		Date of post-spray counts (2010)	Post-spray counts	
				Cv/bud	PM/bud		*Cv*/bud	PM/bud
Yes	Osoyoos (H)	Shiraz	March 24	15.56 ($\pm$ 5.15)	0.22 ($\pm$ 0.15)	April 28	0	0.89 ($\pm$ 0.35)
Yes	Naramata (A)	Merlot	March 10	10.90 ($\pm$ 2.56)	0.10 ($\pm$ 0.10)	April 29	0	0.90 ($\pm$ 0.52)
Yes	Naramata (A)	Cabernet Franc	March 10	13.50 ($\pm$ 4.94)	0.10($\pm$ 0.10)	April 29	0	0.60 ($\pm$ 0.30)
No	Summerland (D)	Gewürztraminer	March 30	27.70 ($\pm$ 5.86)	0	April 21	35.80 ($\pm$ 15.36)	0

[a]Letters correspond to vineyard locations on Figure 1.

It is worth noting that the damaged shoots had lower numbers of predator mites than the undamaged shoots, but the difference was not found to be statistically significant ($F_{1,76}$=3.6261, *P*=0.061). Previous work has shown that preservation of mite predators is important for the management of *C. vitis* (James and Whitney 1993; Bernard *et al.* 2005; Reinert 2006; James 2007). James *et al.* (2002) report that effective biological control of eriophyid mites in Washington vineyards likely depends on a complex of natural enemies in addition to species of predatory phytoseiid mites. Outbreaks of *C. vitis* in Australia and Washington State have been attributed to several causes, including the use of broad-spectrum, persistent insecticides that harm predators. James (2007) suggested that the appearance of *C. vitis* in Washington State might prove advantageous for grapevine biological control programs, as these mites provide an early season food source for predacious mites before *Tetranychid* spider mite species appear later in the season. Additional study is required to determine the mite predator complex in B.C. and to establish their role in the sustainable management of *C. vitis*.

Figure 4. Developing grapevine shoot severely damaged by grape leaf rust mite, *Calepitrimerus vitis* (Nalepa), feeding in early spring (left) compared with an undamaged shoot (right) from the same cultivar block. Note the shortened internodes, brown and distorted leaves and flower buds, and scarring of the stem on the damaged shoot.

In conclusion, our research has demonstrated that reduced spring growth of shoots having deformed leaves that had often been attributed to herbicide or winter damage, post-harvest water stress, thrips, and other maladies is in many cases due to feeding damage from *C. vitis*. Presence of this eriophyid mite under bud scales and on developing shoots has been linked to short shoot syndrome of grapevines, an economically important syndrome of grapes in the Pacific Northwest of the United States (Walton *et al.* 2007; Schreiner *et al.* 2014). We determined that *C. vitis* were distributed widely in the southern interior of B.C. and observed high numbers feeding on buds and tender shoots in spring (Figure 3) that resulted in distortion and stunting of shoots (Figure 4; Table 3). Although growth often improves during the summer, yields will be reduced significantly, as we have shown.

The recommendation to apply sulphur at the woolly bud stage based on bronzing of leaves in late summer is supported by our observations. Evidence for potentially damaging populations of these mites based on bronzing of leaves in late summer is an indication that control measures should be applied the following spring. Although there is no reported damage to grapes from *C. vitis* feeding during the summer, they can be controlled at this time with foliar sprays of miticides (Anonymous 1968; Walton *et al*. 2007; Siquera *et al*. 2016).

With the documented arrival of *C. vitis* to the southern interior of B.C., it is important that growers learn to recognize early signs of severe *C. vitis* infestations (russeting of leaves) that indicate the need for timely and appropriate sulphur sprays during early bud development the following spring. Attempts should also be made to preserve mite predators by avoiding the use of harmful sprays.

ACKNOWLEDGEMENTS

We extend our gratitude to Dr. F. Beaulieu, AAFC Ottawa, and Dr. J. Amrine, West Virginia University, for identification of *C. vitis*. Thank you to M. Weis of AAFC-Summerland RDC for photographs, P. Bowen and B. Estergaard, AAFC-Summerland RDC for use of the map, M. Watson of Constellation Brands International, and other producers for allowing access to their vineyard sites.

Table 3

Assessment in spring[a] of shoot lengths and counts of *Calepitrimerus vitis* [*Cv*], phytophagous thrips, and predatory mites [PM] on four rows of Chardonnay grapes heavily infested with *C. vitis* compared with undamaged rows from the same Okanagan Falls cultivar block was followed by measures in fall[b] of numbers of shoots, grape clusters and cluster lengths (means ± SE).

Vine Row Shoot Status	Spring Shoot Assessment						Pre-harvest Assessment		
	shoot length (cm)	*Cv*/stem	*Cv*/basal leaf	thrips/stem	thrips/basal leaf	PM/shoot	shoots (number)	grape clusters (number)	cluster length (cm)
Undamaged	9.98 (± 0.61)	0	0	0.16 (± 0.06)	0.11 (± 0.06)	0.32 (± 0.10)	8.3 (± 0.38)	10.05 (± 0.66)	10.2 (± 0.57)
Cv-damaged	4.90 (± 0.23)	17.6 (± 2.91)	9.48 (± 2.11)	0.35 (± 0.09)	0.18 (± 0.08)	0.10 (± 0.05)	7.15 (± 0.31)	2.7 (± 0.31)	6.56 (± 0.57)
Significance level[c]	***	***	***	ns	ns	ns	*	***	***

[a]Counts on stems and basal leaves of 20 randomly selected shoots per row on May 18, 2010.
[b]Measures from one cordon arm (1/2 vine) from 20 randomly selected vines per row on September 15, 2010.
[c]Level of significance assessed with a one-way analysis of variance (α=0.05) and indicated as follows: ns, P>0.05; *, P<0.05; **P<0.01; ***P<0.001. All mite count data was transformed before analysis ($\sqrt{(X+0.5)}$).

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